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Research Article

Effects of Orchard-Level Variation and Cultivation Method on the Functional Quality of Indian Jujube (*Ziziphus mauritiana*)

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Abstract

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This study investigated the effects of cultivation method (organic vs. conventional) and orchard-level variation on the functional quality of Indian jujube (*Ziziphus mauritiana*) fruits. A total of 64 fruit samples were collected from four orchards in Taiwan- two organic and two conventional- and analyzed for their antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content. Although organically grown fruits exhibited slightly higher mean levels of antioxidant capacity, phenolic compounds, and flavonoid compounds, linear mixed-effects models indicated that the cultivation method had no statistically significant effect on any functional indicators. In contrast, orchard-level random effects accounted for a substantial proportion of the total variance, particularly for antioxidant capacity, total flavonoids, and vitamin C. Notable differences were observed between orchards within the same cultivation method, suggesting that localized environmental or management conditions may play a stronger role than cultivation system alone in shaping fruit phytochemical profiles. Measured soil nutrient profiles revealed significant differences among orchards, providing plausible explanatory cues for observed phytochemical variation. Pearson correlation analysis further revealed that antioxidant capacity was strongly associated with total phenolic and flavonoid content. In contrast, vitamin C showed no significant correlations with other traits, indicating that phenolic and flavonoid compounds are likely the primary contributors to antioxidant potential in Indian jujube fruits.

Key words: Indian jujube, Functional quality, Cultivation method, Orchard-level variation, Linear mixed-effects model.

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INTRODUCTION

Ziziphus mauritiana Lam., commonly known as Indian jujube, is a member of the Rhamnaceae family and has a long history of cultivation in South Asia and East Africa (O'Brien *et al*. 2023). The species is widely cultivated across tropical and subtropical regions of Asia, including India, Pakistan, China, and Taiwan, where it is regarded as an important economic fruit crop (Koley *et al*. 2016; Duan & Chen 2022). Due to its adaptability to harsh environments and high nutritional value, Indian jujube is also valued in several North and East African countries, where it contributes to food security in resource-limited arid regions (Maruza *et al*. 2017; O'Brien *et al*. 2023).

In Taiwan, Indian jujube- locally known as "zaozi" or "mizao"- is a culturally significant fruit that is widely consumed during the Lunar New Year season. It is associated with prosperity and auspiciousness, making it a popular choice among consumers and a key source of seasonal income for farmers. The crop is cultivated extensively in southern Taiwan, particularly in Kaohsiung, Pingtung, and Tainan, covering approximately 1,700 ha (Agriculture Statistics Inquiry System, https://agrstat.moa.gov.tw/sdweb/public/inquiry/InquireAdvance.aspx). In addition to fresh consumption, Indian jujube is processed into a variety of value-added products, including candied jujube, dried jujube, and jujube juice, thereby enhancing its economic potential.

Indian jujube is known for its sweet flavor and high nutritional value, containing carbohydrates, proteins, dietary fiber, and essential minerals such as potassium, calcium, sodium, phosphorus and magnesium (Nyanga *et al*. 2013; Prakash *et al*. 2021). It is also a rich source of vitamins, including thiamine, riboflavin, niacin, and ascorbic acid (Prakash *et al*. 2021; Shady *et al*. 2022). Of particular interest are its polyphenolic and flavonoid constituents, which have been associated with antioxidant and health-promoting properties (Koley *et al*. 2016; Shady *et al*. 2022). Identified phenolic acids include caffeic acid, chlorogenic acid, ferulic acid, and p-coumaric acid, while flavonoids such as quercetin 3-O-glucoside, epicatechin, and naringenin are also present (Memon *et al*. 2013; Prakash *et al*. 2021; Khanam *et al*. 2025). Furthermore, Indian jujube has been reported to exhibit anti-inflammatory and antidiabetic effects in pharmacological studies (Sangeethapriya & Siddhuraju 2014; Khanam *et al*. 2025), and its high dietary fiber content may support digestive health (Sarkar *et al*. 2022).

With growing demand for food safety and health, organic fruit production has gained increasing attention. Organic agriculture emphasizes ecological sustainability by minimizing the use of synthetic pesticides and fertilizers, and it is often associated with the production of higher-quality and potentially more health-promoting fruits. Several studies have reported that organically grown fruits and vegetables exhibit elevated levels of bioactive compounds, including polyphenols and vitamin C, compared to conventionally grown counterparts (Brandt *et al*. 2011; Średnicka-Tober *et al*. 2020). While the nutritional profile of Indian jujube is known to be influenced by genotype and cultivation environment (Koley *et al*. 2016; Meena *et al*. 2023), a notable lack of research remains in comparing the functional composition of organically and conventionally grown Indian jujube, particularly in subtropical regions like Taiwan. Moreover, orchard-specific environmental and management conditions, such as microclimate, soil fertility, and tree vigor, may also modulate fruit phytochemical expression but are rarely considered in comparative studies (Stracke *et al*. 2009; Rocchetti *et al*. 2022).

This study aims to evaluate how different cultivation systems (organic vs. conventional) and orchard-level variation (such as soil physicochemical properties) influence the functional quality of Indian jujube fruits cultivated in Taiwan. Here, functional quality refers to phytochemical attributes with antioxidant relevance, specifically total phenolic content, total flavonoid content, vitamin C concentration, and overall antioxidant capacity. Through biochemical analyses and linear mixed-effects modeling,

this research provides an integrative perspective on how farming practices and site-specific conditions shape the phytochemical profiles of Indian jujube. The results are expected to inform both scientific understanding and support evidence-based strategies for enhancing the nutritional value of this culturally and economically significant fruit crop.

MATERIALS AND METHODS

Plant materials

This study was conducted at the Kaohsiung Yanchao Long-Term Ecological Research Station in southern Taiwan, which features a tropical monsoon climate (annual rainfall ~1,800 mm, mostly from May to October; average temperature 23–25℃). Fruit samples were collected from four independent orchard blocks, each ≥ 0.2 ha in area, planted with the same variety (*Z. mauritiana* 'Mizao') under either organic or conventional cultivation systems. Detailed orchard information is provided in Appendix 1, and orchard images and locations are shown in Appendix 2. These orchards were designated as follows:

Organic orchards (orchards A and B) are located in Guanshui and Antong, respectively, within the Yanchao District. The orchards are approximately 4 km apart and have been managed organically for over 20 years. The organic production process is certified by Blue Magpie Certifications (Certification No. 1-017-224006, Taipei, Taiwan) and adheres to the Shumei Natural Agriculture philosophy, which prohibits the use of pesticides, chemical fertilizers, and organic manures. Trees are grown under 16-mesh net house conditions with single-layer horizontal trellising, spaced 6 m × 6 m, and irrigated using well water.

Conventional orchards (Orchards C and D) are located in the Guanshui section of Yanchao District. The orchards occupy adjacent cadastral parcels and have been managed under conventional cultivation practices for over 10 years. They use a 6 m × 6 m planting grid and cultivate under 16-mesh nylon netting. Fertilization involves the annual application of NPK fertilizers a 4:2:5 ratio, with 1,500 g applied per tree. Pest and disease control involves the timely application of registered agrochemicals: 10% fenpyroximate suspension concentrate (SC) at 2,000× dilution (1,000 L ha^{-1}) for mite control; 40% kresoxim-methyl wettable powder (WP) at 1,000× dilution (2,000 L ha^{-1}) during the fruiting stage for powdery mildew control; and 23% azoxystrobin SC at 3,000× dilution (2,000 L ha^{-1}) during the fruiting stage for anthracnose control.

Across all orchards, only trees aged ≥ 10 years showing healthy growth without visible pest or disease damage were selected. For each orchard, a total of 16 fruits per orchard were randomly harvested from mid-canopy positions at commercial maturity (~80% ripeness), as identified by yellow-green skin with slight softening (Fig. 1) (Guo *et al*. 2022). The fruits were stored at ambient temperature for subsequent phytochemical and antioxidant analyses.

Sample preparation for phytochemical analyses

Approximately 50 g of fresh Indian jujube fruit was homogenized with 200 mL of distilled water using a commercial blender (JVE-1758W, CookPower, New Taipei, Taiwan) to produce a uniform juice. A 20 mL aliquot of the juice was then mixed with 20 mL of methanol and subjected to ultrasonic extraction. Ultrasonic extraction was conducted using an ultrasonic bath (DC400H, DELTA, New Taipei, Taiwan) operating at 40 kHz and 400 W for 20 min at room temperature. After extraction, the mixture was

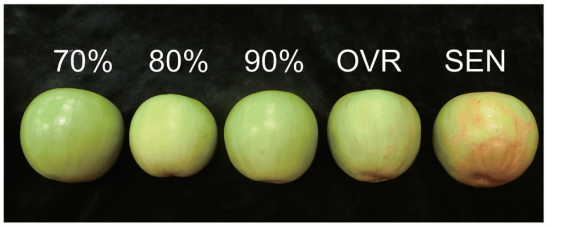


Fig. 1. Indian jujube fruit maturity stages. From left to right: 70% mature, 80% mature (commercial standard), 90% mature, overripe (OVR), and senescence (SEN).

centrifuged at 2,400 × *g* for 10 min using a centrifuge (Model 4000, KUBOTA, Osaka, Japan). The resulting supernatant was collected and used to determine the antioxidant capacity, total phenolic content, and total flavonoid content.

Antioxidant capacity

Antioxidant capacity was evaluated using the ABTS radical cation decolorization assay, as described by Thaipong *et al*. (2006), with slight modifications. The $ABTS^{\bullet+}$ solution was prepared by reacting 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) (A3219, Sigma-Aldrich, Saint Louis, MO, USA) with 30 mM ammonium persulfate (101200, Merck, Darmstadt, Germany) at a 10:1 (v:v) ratio and allowing the mixture to stand in the dark at room temperature for 1.5 h to generate the radical cation. Before the assay, the $ABTS^{\bullet+}$ solution was diluted with distilled water to obtain an absorbance of 1.0–1.5 at 734 nm.

For the measurement, 180 µL of diluted $ABTS^{\bullet+}$ solution was mixed with 20 µL of the sample extract or distilled water (blank) in a 96-well microplate. The absorbance was measured at 734 nm at room temperature using a microplate reader (SpectraMax250, Molecular Devices, San Jose, CA, USA). Trolox (0–800 µM) was used to generate the standard calibration curve. Results were expressed as Trolox equivalent antioxidant capacity per gram of fresh weight (mg TE g^{-1}).

Total phenolic content: The total phenolic content of the jujube extracts was determined using the Folin-Ciocalteu colorimetric method, as described by Noreen *et al*. (2017), with minor modifications. Briefly, 50 µL of sample extract was mixed with 50 µL of 0.5N Folin-Ciocalteu reagent (109001, Merck, Darmstadt, Germany) in a 96-well microplate. After 5 min, 100 µL of a 20% sodium carbonate solution (31432, Honeywell, Offenbach am Main, Germany) was added, and the mixture was incubated at room temperature in the dark for 5 min. The absorbance was measured at 750 nm using a microplate reader. Gallic acid (410862500, Acros Organics, Geel, Belgium) was used as a standard to construct the calibration curve (0–100 µg mL^{-1}), and results were expressed as milligrams of gallic acid equivalents per gram of fresh weight (mg GAE g^{-1}).

Total flavonoid content: The total flavonoid content of the jujube extracts was determined following the colorimetric method described by Do *et al*. (2014), with slight modifications. Briefly, 100 µL of the sample extract was mixed with 25 µL of 2% sodium nitrite ($NaNO_2$) solution and allowed to react for 6 min at room temperature. Then, 30 µL of 10% aluminum chloride ($AlCl_3$) solution was added, followed by incubation for another 6 min. Subsequently, 200 µL of 1 M sodium hydroxide (NaOH) was added, and the final volume was adjusted to 1 mL with distilled water. The absorbance was measured at 510 nm using a microplate reader. Rutin was used to generate the standard calibration curve (0–500 µg mL^{-1}), and results were expressed as milligrams of rutin equivalents per gram of fresh weight (mg RUE g^{-1}).

Vitamin C content

The vitamin C content of fresh jujube fruits was determined using an RQflex® 20 reflectometer (Merck, Darmstadt, Germany) in conjunction with the corresponding Ascorbic Acid Test strips (Product No. 116981, Merck, Darmstadt, Germany), following the manufacturer's instructions. Fresh fruits were finely grated using a stainless-steel grater to obtain juice. A small volume of the juice was applied directly onto the reaction zone of the test strip, which was then inserted into the reflectometer. The instrument automatically recorded the change in reflectance intensity and calculated the vitamin C content by referencing an internal calibration curve. Results were expressed as milligrams of vitamin C per 100 mL of juice (mg 100 mL^{-1}).

Soil fertility analysis

Soil samples were collected before jujube fruit harvest (December) from each orchard. For each orchard, 5 random sampling points were selected. At each point, soil was taken from 3 depths: surface (0–15 cm), subsurface (15–30

cm), and bottom (30–45 cm). The samples from the same orchard were thoroughly mixed and air-dried for 5–7 d at ambient temperature. After drying, the soil was gently crushed to break up clods, passed through a 1 mm sieve, and stored for analysis. Total nitrogen, total carbon, and total organic carbon (TOC) were determined by weighing 0.3 g of soil and analyzing it with a Soli TOC cube analyzer (Elementar, Langenselbold, Germany). Soil organic matter content was calculated as TOC × 1.724, following the Van Bemmelen factor assumption that organic matter contains 58% carbon (Nelson & Sommers 1996). Available phosphorus, exchangeable potassium, exchangeable calcium, and exchangeable magnesium were determined by weighing 2.0 g of soil into a 50 mL conical flask, adding 20 mL of Mehlich No. 3 extractant, and shaking at 200 rpm for 5 min. The suspension was filtered, and the filtrate analyzed using an inductively coupled plasma–optical emission spectrometer (iCAP 7000 Series, Thermo Fisher Scientific, Waltham, MA, USA).

Statistical analysis

To appropriately model the hierarchical structure of the data and account for site-level heterogeneity, we employed linear mixed-effects models (LMMs), constructed using the lme4 package (version 1.1.37) in R (version 4.3.2, R Core Team, Austria). This approach was necessary due to the nested experimental design, where orchard identity was nested within cultivation method (organic vs. conventional). LMMs enable the partitioning of variance into fixed and random components, providing unbiased estimates of treatment effects while adjusting for the non-independence of observations collected within the same orchard. Cultivation method was treated as a fixed effect, and orchard identity (nested within cultivation method) was included as a random effect. Separate models were constructed for each fruit quality trait: antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content.

To assess fixed effects (cultivation method), we computed estimated marginal means (EMMs) for each cultivation method using the emmeans package (R package version 1.6.1), which adjusts for the structure of the random effects. The statistical significance of fixed effects was evaluated using Type II Wald χ^2 tests, implemented in the car package. The contribution of the random effect (orchard) was tested using likelihood ratio tests by comparing full and reduced models (excluding the random term) via the anova function.

To evaluate model explanatory power, marginal R^2 (R^2m) and conditional R^2 (R^2c) were calculated using the performance package, following the method of Nakagawa & Schielzeth (2013). R^2m quantifies variance explained by fixed effects alone, while R^2c accounts for both fixed and random effects.

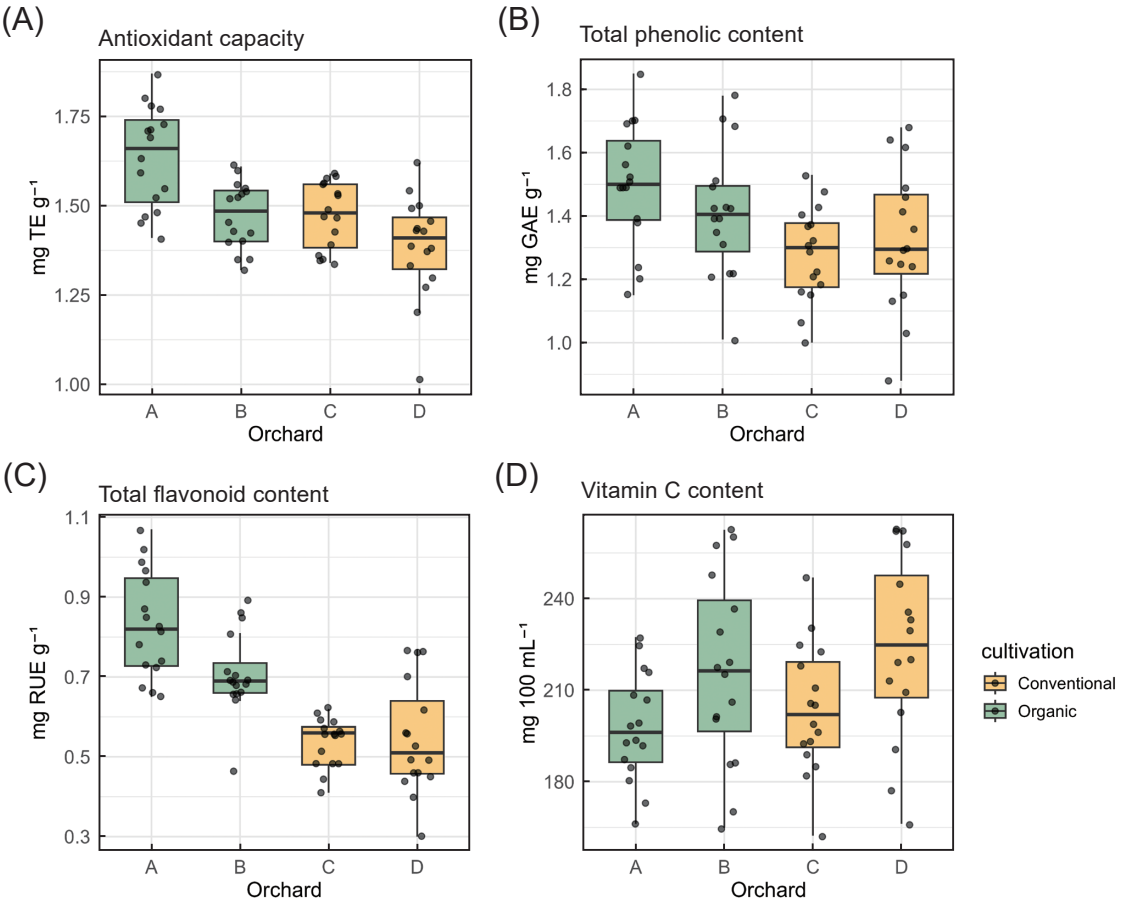
To visualize orchard-level variation within cultivation methods, boxplots were generated using the ggplot2 package. Finally, Pearson's correlation coefficients were calculated using the base R cor function to assess associations between antioxidant capacity and the three phytochemical traits. Statistical significance was determined at $P < 0.05$.

RESULTS

Descriptive overview of functional quality indicators

A total of 64 Indian jujube fruit samples were collected from four orchards, each representing one of two cultivation methods- conventional (orchards C and D) or organic (orchards A and B). Each orchard contributed 16 fruit samples. Four functional indicators were evaluated: antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content.

Descriptive statistics for all variables across the four orchards are summarized in Fig. 2. In terms of antioxidant capacity, Orchard A (organic) exhibited the highest mean value (1.64 ± 0.14 mg TE g^{-1}), followed by Orchard C



(E)

Cultivation method	Orchard	Antioxidant capacity (mg TE g^{-1})	Total phenolic content (mg GAE g^{-1})	Total flavonoid content (mg RUE g^{-1})	Vitamin C content (mg 100 mL^{-1})
Organic	A	1.64 ± 0.14	1.50 ± 0.19	0.83 ± 0.13	197.9 ± 17.8
Organic	B	1.47 ± 0.09	1.41 ± 0.20	0.71 ± 0.11	216.3 ± 31.1
Conventional	C	1.47 ± 0.09	1.28 ± 0.15	0.54 ± 0.06	203.8 ± 21.2
Conventional	D	1.38 ± 0.14	1.32 ± 0.22	0.55 ± 0.14	224.1 ± 30.2

Fig. 2. (A–D) Boxplots of antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content in Indian jujube fruits from four orchards (n = 16 per orchard). (E) Descriptive statistics (mean ± *SD*) for each functional component under different cultivation methods and orchards. Orchards A and B were organic; orchards C and D were conventional. Boxplots show the interquartile range (IQR, box), median (horizontal line), whiskers (1.5 × IQR), and individual samples (jittered points). Units: antioxidant capacity (mg TE g^{-1}), total phenolic content (mg GAE g^{-1}), total flavonoid content (mg RUE g^{-1}), vitamin C (mg 100 mL^{-1}).

(1.47 ± 0.09 mg TE g^{-1}) and Orchard B (1.47 ± 0.09 mg TE g^{-1}), with Orchard D (conventional) showing the lowest value (1.38 ± 0.14 mg TE g^{-1}).

For phenolic content, Orchard A again showed the highest mean value (1.50 ± 0.19 mg GAE g^{-1}), followed by Orchard B (1.41 ± 0.20

mg GAE g^{-1}), Orchard D (1.32 ± 0.22 mg GAE g^{-1}), and Orchard C (1.28 ± 0.15 mg GAE g^{-1}). Total flavonoid content showed a more pronounced difference between cultivation types, with organic orchards A and B reporting higher mean values (0.83 ± 0.13 and 0.71 ± 0.10 mg RUE g^{-1}, respectively) compared to conventional orchards C and D (0.54 ± 0.06 and 0.55 ± 0.14 mg RUE g^{-1}, respectively) (Fig. 2).

The vitamin C content varied across orchards without a clear pattern associated with cultivation method. Orchard D exhibited the highest mean value (224.1 ± 30.2 mg 100 mL^{-1}), followed by Orchard B (216.3 ± 31.1 mg 100 mL^{-1}), Orchard C (203.8 ± 21.2 mg 100 mL^{-1}), and Orchard A (197.9 ± 17.8 mg 100 mL^{-1}) (Fig. 2). These results suggest that, while some functional components, such as flavonoids, appear more responsive to cultivation method, orchard-specific factors may influence like vitamin C and others.

Effect of cultivation method on functional quality of indian jujube fruits

The influence of cultivation method on the functional quality of Indian jujube fruits was assessed using linear mixed-effects models, with orchard nested within cultivation method as a random effect. As shown in Table 1, the fixed effect of cultivation method was not statistically significant for antioxidant capacity (P = 0.313), total phenolic content (P = 0.225), or vitamin C content (P = 0.628). However, the difference in total flavonoid content between organic and conventional samples (P = 0.053) approached statistical significance (P = 0.05).

Estimated marginal means were also illustrated in Table 1. Although without significance, organic fruits showed higher mean values than conventional fruits for antioxidant capacity (1.55 vs. 1.43 mg TE g^{-1}), total phenolic content (1.45 vs. 1.30 mg GAE g^{-1}), and total flavonoid content (0.77 vs. 0.54 mg RUE g^{-1}). In contrast, the mean vitamin C content of organic fruits was slightly lower than that of conventional fruits (207.1 vs. 213.9 mg 100 mL^{-1}), and this difference was also not statistically significant.

The marginal R^2 values from Nakagawa's method ranged from 0.077 to 0.267 across models, suggesting that cultivation method alone explained a modest proportion of the total variance. Conditional R^2 values were higher (0.338 to 0.443), indicating that orchard-level random effects also contributed meaningfully to the observed variation (Table 1).

Variation at the orchard level in phytochemical profiles

The random effect using the linear mixed-effects models is shown in Table 2. The variance components for orchard-level effects were found to be statistically significant for antioxidant capacity (P < 0.001), total flavonoid content (P = 0.013), and vitamin C content (P = 0.015). Conversely, total phenolic content did not exhibit a significant random effect at the orchard level (P = 0.347).

Table 1. Estimated marginal means and statistical significance of functional compounds in Indian jujube fruits under different cultivation methods.

Response variable	Organic (EMM ± *SE*)[z]	Conventional (EMM ± *SE*)	*P*-value	Marginal R^{2y}	Conditional R^{2x}
Antioxidant capacity (μmol TE g^{-1})	1.55 ± 0.08	1.43 ± 0.08	0.313	0.149	0.443
Total phenolic content (mg GAE g^{-1})	1.45 ± 0.10	1.30 ± 0.10	0.225	0.267	0.338
Total flavonoid content (mg RUE g^{-1})	0.77 ± 0.06	0.54 ± 0.06	0.053	0.182	0.360
Vitamin C content (mg 100 mL^{-1})	207.1 ± 9.6	213.9 ± 9.6	0.628	0.077	0.410

[z] Estimated Marginal Means (EMM) ± Standard Error (*SE*) derived from linear mixed-effects models.
[y] Marginal R^2 represents the proportion of variance explained by the fixed effects (cultivation method) alone.
[x] Conditional R^2 represents the proportion of variance explained by both fixed effects and random effects (orchard). R^2 values were calculated using Nakagawa's method.

Soil physicochemical properties of the orchards

Soil physicochemical properties of the four orchards are summarized in Table 3. Soil pH values were uniformly alkaline, ranging from 7.91 to 7.99. Organic matter content was highest in Orchard B (2.961%), followed closely by Orchard A (2.901%), and was lower in Orchard C (2.137%) and lowest in Orchard D (1.995%). Total carbon content was also highest in Orchard A (1.874%), followed by Orchard B (1.864%), Orchard C (1.390%), and Orchard D (1.255%). Total nitrogen content was highest in Orchard A (0.255%), followed by Orchard B (0.173%), Orchard D (0.132%), and Orchard C (0.130%). Available phosphorus varied markedly, with Orchard C (923.9 mg kg^{-1}) and Orchard A (618.8 mg kg^{-1}) showing much higher concentrations than Orchard B (51.6 mg kg^{-1}) and Orchard D (39.1 mg kg^{-1}). Exchangeable potassium was highest in Orchard C (643.6 mg kg^{-1}) and lowest in Orchard D (120.1 mg kg^{-1}). Exchangeable calcium ranged from 2,424.8 mg kg^{-1} in Orchard D to 3,703.1 mg kg^{-1} in Orchard A, whereas exchangeable magnesium was highest in Orchard C (1,030.8 mg kg^{-1}) and lowest in Orchard B (572.4 mg kg^{-1}).

Correlations among functional compounds

Pearson correlation analyses were performed to investigate relationships among antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content across all samples (n = 64). As shown in Table 4, antioxidant capacity was strongly and positively correlated with total phenolic content (r = 0.85, P < 0.001) and total flavonoid content (r = 0.79, P < 0.001). Total phenolic and flavonoid contents were also significantly correlated (r = 0.76, P < 0.001). In contrast, vitamin C content showed no significant correlation with antioxidant capacity (r = 0.12, P = 0.35), total phenolic content (r = 0.08, P = 0.48), or flavonoid content (r = 0.10, P = 0.42). These results indicate that phenolic and flavonoid compounds likely contribute substantially to the antioxidant capacity of Indian jujube fruits. In contrast, vitamin C may play a less prom-

Table 2. Random effects of orchard nested within cultivation method on antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content in Indian jujube fruits (n = 64).

Response variable	Random effect variance (σ^2)[z]	P-value[y]
Antioxidant capacity	0.0077	< 0.001
Total phenolic content	0.0004	0.347
Total flavonoid content	0.0021	0.013
Vitamin C content	25.43	0.015

[z] Random effects are estimated using the restricted maximum likelihood method in linear mixed-effects models.
[y] P-values are based on likelihood ratio tests comparing models with and without orchard-level random intercept.

Table 3. Soil physicochemical properties in four orchards.

Orchard	pH (1 : 1 soil : water)	Organic matter (%)	Total carbon (%)	Total nitrogen (%)	Available phosphorus (mg kg^{-1})	Exchangeable potassium (mg kg^{-1})	Exchangeable calcium (mg kg^{-1})	Exchangeable magnesium (mg kg^{-1})
A	7.97	2.901	1.874	0.255	618.8	340.9	3,703.1	966.9
B	7.91	2.961	1.864	0.173	51.6	158.5	3,378.8	572.4
C	7.91	2.137	1.390	0.130	923.9	643.6	3,611.3	1,030.8
D	7.99	1.995	1.255	0.132	39.1	120.1	2,424.8	702.2

Table 4. Pearson correlation coefficients among antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content in Indian jujube fruits (n = 64).

Response variables	Antioxidant capacity	Total phenolic content	Total flavonoid content	Vitamin C content
Antioxidant capacity	1			
Total phenolic content	0.85 ($P < 0.001$)[z]	1		
Total flavonoid content	0.79 ($P < 0.001$)	0.76 ($P < 0.001$)	1	
Vitamin C content	0.12 ($P = 0.35$)[y]	0.08 ($P = 0.48$)	0.10 ($P = 0.42$)	1

[z] Correlation significant at $P < 0.001$.
[y] Correlation not statistically significant ($P > 0.05$).

inent role in determining antioxidant activity under the tested conditions.

DISCUSSION

Influence of cultivation method on functional quality of indian jujube fruits

In this study, cultivation method (organic vs. conventional) did not exert a statistically significant effect on any of the measured quality traits, including antioxidant capacity, total phenolic content, total flavonoid content, and vitamin C content (Table 1). While fruits from organic orchards tended to exhibit slightly higher mean levels, for example, antioxidant capacity and flavonoid content, these differences were modest. Only total flavonoid content approached statistical significance (P = 0.053), a value close to the conventional threshold (P = 0.05) (Table 1). The tendency for flavonoid content is likely linked to typical organic practices, where reduced synthetic inputs and increased exposure to environmental stressors stimulate secondary metabolite synthesis (Jan *et al*. 2021; Khan *et al*. 2025). Specifically, practices such as limited pesticide use and lower nitrogen availability can heighten plant defense responses, leading to the upregulation of phenylpropanoid pathways responsible for polyphenol biosynthesis and the accumulation of enhanced antioxidant compounds as part of the plant's adaptive strategy (Stewart *et al*. 2001; Krey *et al*. 2020). Nevertheless, under the specific conditions examined in this study, cultivation system alone did not appear to be a dominant factor shaping fruit phytochemical profiles.

Orchard-Level variation in phytochemical composition and its implications

The linear mixed-effects models effectively captured the heterogeneity at the orchard level. The random effects of orchard nested within cultivation method accounted for a substantial proportion of the total variance, particularly for antioxidant capacity ($P < 0.001$), total flavonoid content (P = 0.013), and vitamin C content (P = 0.015) (Table 2). In this study, orchard-level variation is defined as the cumulative influence of site-specific environmental conditions and managerial practices that are not explicitly represented by the binary classification of cultivation methods (organic vs. conventional). These factors include soil physicochemical properties, orchard history, microclimatic differences, and other unmeasured field-specific variables. Such factors can modulate plant physiological processes and influence the accumulation of phytochemicals (Stracke *et al*. 2009; Rocchetti *et al*. 2022). In essence, orchard-level variation reflects the integrated field context in which a crop is grown, and its effects can even surpass those attributable to cultivation system categories.

Among the potential contributors to orchard-level variation identified above, measured soil nutrient profiles (Table 3) may offer plausible explanatory cues. For example, the higher antioxidant capacity and total flavonoid content observed in organic Orchard A relative to Orchard B (Fig. 2) may be attributable to the combined effects of several edaphic factors. While the two orchards exhibited similar organic matter and total carbon levels, Orchard A had

higher concentrations of nitrogen, phosphorus, potassium, and magnesium (Table 3). From a physiological perspective, a balanced supply of macronutrients and magnesium enhances chlorophyll biosynthesis and photosynthetic efficiency, thereby increasing the flux of fixed carbon toward sink organs (Farhat *et al*. 2016; Wang *et al*. 2024). This expanded carbon pool supports the energetically demanding pathways for flavonoid biosynthesis (Deng *et al*. 2019), potentially explaining the superior flavonoid content in Orchard A. Such relationships between nutrient and secondary metabolism linkages have been consistently reported in fruit crops (Pott *et al*. 2019), reinforcing nutrient balance as one of the reliable drivers of orchard-level differences.

In contrast, the conventional group highlights a different facet of orchard-level variation. Orchard C displayed higher soil organic matter and multiple macronutrients than Orchard D (Table 3). Yet, the two orchards showed no appreciable difference in phytochemical metrics (Fig. 2). This finding aligns with the hypothesis that excessive nutrient supply may not translate into proportional quality gains in conventional systems (Kim *et al*. 2020; Milošević *et al*. 2022). Possible explanations include the lack of a robust soil microbial network to facilitate nutrient transformation and uptake, which is often more developed in long-term organic systems (Rodríguez-Ortiz *et al*. 2022; Wang *et al*. 2022). This observation highlights that nutrient abundance alone may not be sufficient. The bioavailability of nutrients and the efficiency of their physiological integration are equally critical.

Although we lack direct measurements of canopy light or temperature profiles, numerous studies in perennial fruit crops suggest that site-specific microclimate and light likely interact with soil nutrient status to modulate phytochemical outcomes (Vaneková *et al*. 2020; Han *et al*. 2023). Future research should integrate more environmental monitoring, such as canopy light mapping, leaf water potential tracking, and soil microbial community profiling, to better understand how microclimate-nutrient interactions drive phytochemical variation across orchards. Taken together, our results reinforce the conclusion that orchard-level variation can exert a stronger influence on the functional quality of Indian jujube than cultivation method alone.

Relationships between functional compounds and antioxidant capacity

Antioxidant capacity was strongly and positively correlated with both total phenolic content (r = 0.81, P < 0.001) and total flavonoid content (r = 0.78, P < 0.001), indicating that these two classes of polyphenolic compounds are the principal contributors to the antioxidant potential of Indian jujube fruits. This finding is consistent with the established role of phenolics and flavonoids as major radical scavengers in plant-derived foods (Mustafa *et al*. 2010; Muflihah *et al*. 2021). Previous studies on *Ziziphus* species have likewise found phenolic content to be the dominant determinant of antioxidant activity. For instance, Zhang *et al*. (2010) reported a strong association between phenolic concentration and antioxidant potential in Chinese jujube extracts.

In contrast, vitamin C content showed no significant correlation with antioxidant capacity (r = -0.06, P = 0.61), nor with total phenolic content (r = 0.08, P = 0.48) or flavonoid content (r = 0.10, P = 0.42). This suggests that vitamin C plays a comparatively minor role in the antioxidant capacity of Indian jujube under the conditions studied. Although vitamin C is recognized as an important antioxidant, its contribution can vary significantly across species, cultivars, and environmental contexts (Zhang *et al*. 2010; Wojdyło *et al*. 2016).

From an application perspective, these findings suggest that enhancing the functional quality of Indian jujube may be more effectively achieved by promoting polyphenol biosynthesis through targeted stress induction or optimized agronomic practices, rather than by focusing solely on increasing vitamin C levels. Breeding efforts or orchard management strategies that

select for or enhance phenolic and flavonoid accumulation may therefore offer greater nutritional benefits and functional consistency.

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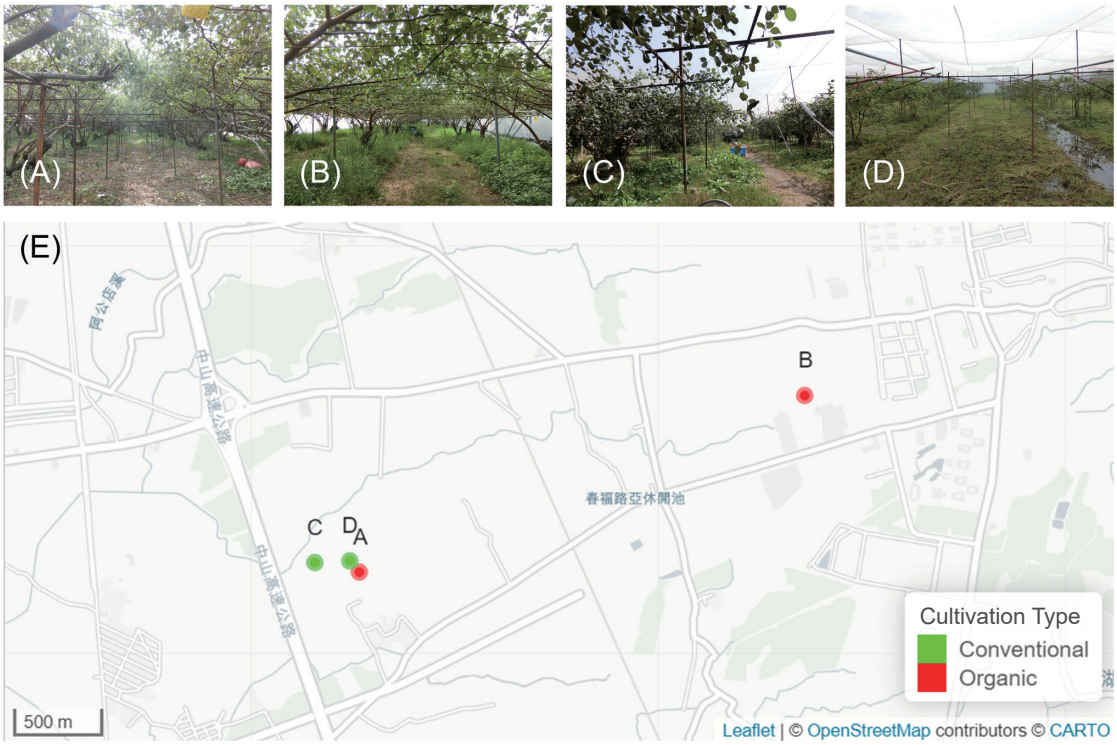
Appendix 1. Geographic location and cadastral information of the four orchards used in this study, including cultivation type, area, cadastral parcel numbers, and global positioning system (GPS) coordinates.

Orchard	Cultivation type	Area (hectare)	Location (cadastral parcels)	GPS
A	Organic	0.4	Guanshui 293, 293-1, 293-2	22.7802,120.3281
B	Organic	0.25	Antong 0705-0000	22.7890,120.3521
C	Conventional	0.4	Guanshui 300, 300-1, 300-2	22.7807,120.3257
D	Conventional	0.3	Guanshui 0283	22.7808,120.3276



Appendix 2. (A–D) Photographs of the four Indian jujube orchards at the Kaohsiung Yanchao Long-Term Ecological Research Station. Orchards A and B were managed organically, while C and D were under conventional cultivation. (E) Location map of the orchards, generated with the R package leaflet.

果園層級變異與栽培方式對印度棗 (*Ziziphus mauritiana*) 機能品質之影響

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摘要

洪子桓、張家華、陳秋樺、陳素漪、徐敏記、郭展宏。2025。果園層級變異與栽培方式對印度棗 (*Ziziphus mauritiana*) 機能品質之影響。台灣農業研究 74(4):525–539。

本研究探討栽培方式 (有機與慣行) 與果園層級變異對印度棗 (*Ziziphus mauritiana*) 果實機能品質的影響。研究從臺灣 4 個果園 (2 個有機果園與 2 個傳統果園) 中採集共 64 個果實樣本，分析其抗氧化力、總酚含量、總黃酮含量及維生素 C 含量。雖然有機果實在抗氧化力、酚類及黃酮類化合物含量上的平均值略高，但線性混合效應模型顯示栽培方式對各項機能指標無顯著統計影響。相較之下，果園層級的隨機效應解釋了總變異中的重要比例，尤其是在抗氧化力、總黃酮及維生素 C 方面。同一栽培方式下的不同果園間，亦觀察到顯著差異，顯示局部環境或管理條件可能比栽培系統本身對果實化學特徵方面具有更大的影響。果園間存在明顯的土壤養分差異，為觀察到的植化素變異提供了合理解釋線索。皮爾森相關性分析顯示抗氧化力與總酚及總黃酮含量呈顯著正相關，而維生素 C 則與其他指標無顯著相關，推測酚類與黃酮類化合物可能是印度棗抗氧化潛力的主要貢獻者。

關鍵詞：印度棗、機能品質、栽培方式、果園層級變異、線性混合效應模型。

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